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Functional Ecological Gene Networks to reveal the changes among microbial interactions under elevated carbon dioxide conditions

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Abstract

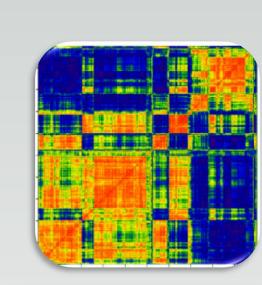
Biodiversity and its responses to environmental changes is a central issue in ecology, and for society. Almost all microbial biodiversity researches focus on species richness and abundance but ignore the interactions among different microbial species/populations. However, determining the interactions and their relationships to environmental changes in microbial communities is a grand challenge, primarily due to the lack of information on the network structure among different microbial species/populations. Here, a novel random matrix theory (RMT)-based conceptual framework for identifying functional ecological gene networks (fEGNs) is developed with the high throughput functional gene array hybridization data from the grassland microbial communities in a long-term FACE (Free Air CO₂) Enrichment) experiment. Both fEGNs under elevated CO₂ (eCO₂) and ambient CO₂ (aCO₂) possessed general characteristics of many complex systems such as scale-free, small-world, modular and hierarchical. However, the topological structure of the fEGNs is distinctly different between eCO₂ and aCO₂, suggesting that eCO₂ dramatically altered the interactions among different microbial functional groups/populations. In addition, the changes in network structure were significantly correlated with soil carbon and nitrogen dynamics, and plant productivity, indicating the potential importance of network interactions in ecosystem functioning. Elucidating network interactions in microbial communities and their responses to environmental changes are fundamentally important for research in microbial ecology, systems microbiology, and global change.

Network construction

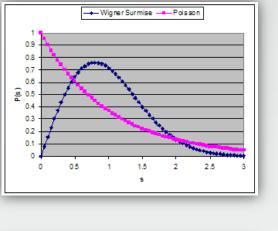
High throughput metagenomics data: *e.g.* GeoChip, sequencing



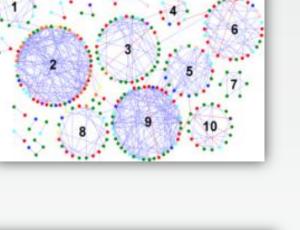
Pair-wise similarity of gene abundance across different samples: e.g. Pearson correlation



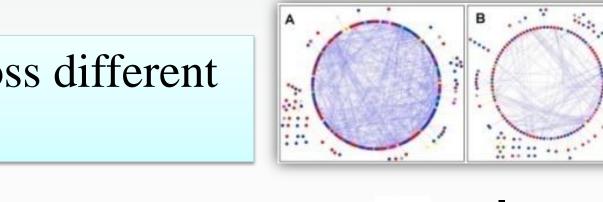
Determination of adjacency matrix by RMT-based approach



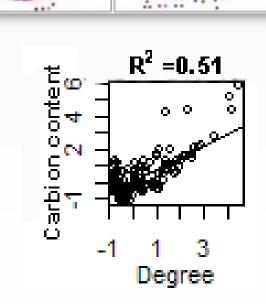
Network characterization and module detection



Network comparison across different samples



Association of network properties to ecological functional traits



| oxidation

Main results | The column of the column of

Fig. 1. Functional genes distributions of nodes in the network under eCO₂ (blue) and aCO₂ (red). The distribution of nodes varies substantially among different gene categories.

Table 1. Summary of the network complexity of represented individual functional genes involved in C, N, P and S cyclings.

Gene Category	Functional group	Selected enzyme/gene names	Number of Shared nodes (%)	aCO_2			eCO_2		
				Number of all nodes	Average degree in shared nodes	Shannon index of degree	Number of all nodes	Average degree in shared nodes	Shannon index of degree
All enzymes/genes			129(43%)	184	3.75	4.879	245	9.11	5.027
Carbon cycling	Carbon degradation	Endochitinase (chi)	8 (57%)	10	3.25	1.885	12	8.25	1.918
		Endoglucanase (bcsG)	0 (0%)	0	0.00	0	2	0.00	1
		exochitinase (chi36)	1 (100%)	1	1.00	0	1	1.00	0
		Exoglucanase (exg)	1 (50%)	1	8.00	0	3	2.50	0.868
		Lignin peroxidase (lip)	0 (0%)	2	0.00	0.305	3	0.00	0.898
		Manganese peroxidase (mnp)	4 (100%)	4	4.25	1.202	4	11.00	1.121
		Pectinase (pglA)	2 (67%)	2	1.00	0.693	3	4.00	0.684
		Phenol oxidase (phox)	4 (36%)	7	2.75	1.681	8	2.00	1.955
		Xylanase (xyn)	5 (38%)	9	3.80	1.687	9	8.00	1.867
	Carbon fixation	Carbon monoxide dehydrogenase (<i>CODH</i>)	2 (40%)	4	4.50	1.137	3	4.50	1.028
		Tetrahydrofolate formylase (<i>FTHFS</i>)	0 (0%)	0	0.00	0	1	0.00	1
		Propionyl-CoA carboxylase (pcc)	9 (36%)	11	2.11	2.224	23	10.56	2.706
		Rubisco (rbcL)	3 (23%)	6	7.67	1.523	10	11.00	1.872
	Methane metabolism	Methyl coenzyme M reductase (<i>mcrA</i>)	0 (0%)	0	0.00	0	2	0.00	0.562
		Methane monooxygenase (pmoA)	1 (14%)	2	3.00	0.562	6	1.00	1.586
Nitrogen cycling	N fixation	Nitrogenase reductase (nifH)	27 (55%)	32	3.07	3.206	44	9.74	3.292
	Denitrification	Nitrite reductase (<i>nirK</i>)	6 (27%)	15	4.00	2.402	13	6.50	2.186
		Nitrite reductase (nirS)	8 (57%)	8	2.88	1.942	14	6.50	2.258
	Dissimilatory N reduction	c-type cytochrome nitrite reductase (<i>nrfA</i>)	4 (57%)	4	7.25	1.356	7	12.25	1.006
Phosphorus cycling		Polyphosphate kinase (ppk)	5 (50%)	7	4.20	1.769	8	11.80	1.671
		Exopolyphosphatase (ppx)	12 (48%)	19	4.83	2.621	18	11.25	2.363
Sulphur	sulfite reductase	Dissimilatory sulfite reductase (<i>dsrA</i>)	13 (34%)	22	5.08	2.66	29	11.54	2.905
	Sulphur oxidation	Sulfite oxidase (sox)	14 (54%)	18	3.00	2.637	22	8.50	2.743

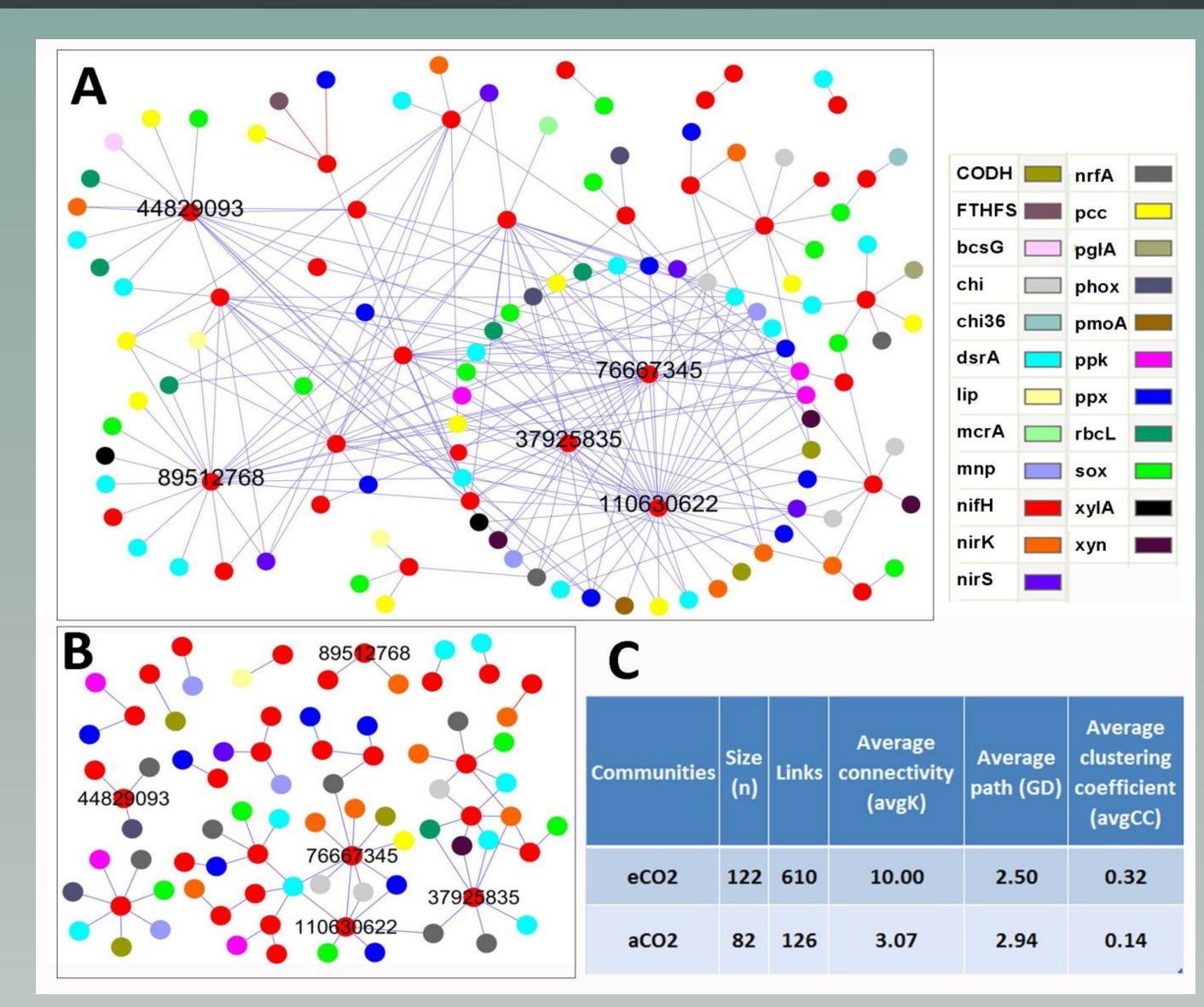


Fig. 2. Network interactions of microorganisms containing *nifH* **genes under eCO₂ (A) and aCO₂ (B).** Microorganisms containing *nifH* genes formed complex network interactions with other functional groups, and some *nifH*-containing populations serve as central hubs in this community. Only shared *nifH* nodes, and their nearest neighbors in the network were shown here. Differences in the colors of the nodes indicate the functional genes in microbial functional groups. The numbers in the nodes represent the GenBank protein IDs to differentiate different *nifH* genes because most of them represent uncultivated microorganisms.

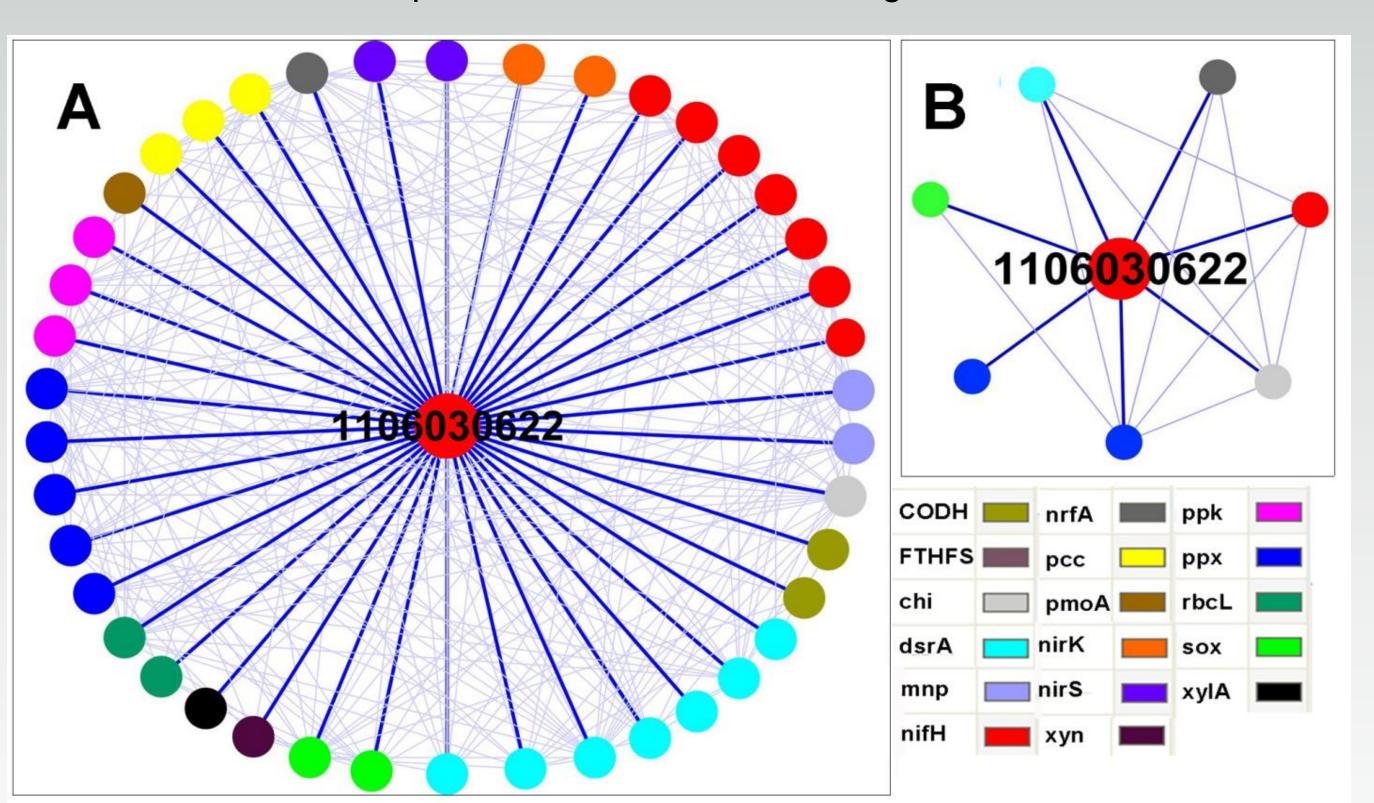


Fig. 3. Network interactions of a *nifH* **hub under both eCO₂** and aCO₂. The *nifH*-containing uncultivated microorganism had intensive positive interactions with many functional groups of diverse phylogenetic origins under eCO₂ (A), but very simple interactions with other functional groups under aCO₂ (B). Only this *nifH* gene node (110630622) and its nearest neighbors are shown. The direct interactions with this nifH gene were labeled with thick lines whereas the indirect interactions were marked with thin lines.

ACKNOWLEDGEMENTS

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